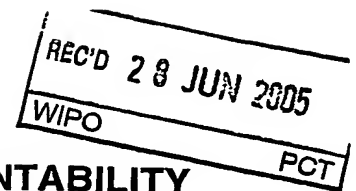


## PATENT COOPERATION TREATY

## PCT

INTERNATIONAL PRELIMINARY REPORT ON PATENTABILITY  
(Chapter II of the Patent Cooperation Treaty)

(PCT Article 36 and Rule 70)





Applicant's or agent's file reference 2M/2AY03/7	<b>FOR FURTHER ACTION</b> See Form PCT/PEA/416	
International application No. PCT/NL2004/000244	International filing date (day/month/year) 13.04.2004	Priority date (day/month/year) 14.04.2003
International Patent Classification (IPC) or national classification and IPC A01H1/04		
Applicant BEJO ZADEN B.V. et al.		

1. This report is the international preliminary examination report, established by this International Preliminary Examining Authority under Article 35 and transmitted to the applicant according to Article 36.
2. This REPORT consists of a total of 8 sheets, including this cover sheet.
3. This report is also accompanied by ANNEXES, comprising:
  - a. ☐ sent to the applicant and to the International Bureau a total of sheets, as follows:
    - ☐ sheets of the description, claims and/or drawings which have been amended and are the basis of this report and/or sheets containing rectifications authorized by this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions).
    - ☐ sheets which supersede earlier sheets, but which this Authority considers contain an amendment that goes beyond the disclosure in the international application as filed, as indicated in item 4 of Box No. I and the Supplemental Box.
  - b. ☐ (sent to the International Bureau only) a total of (indicate type and number of electronic carrier(s)) , containing a sequence listing and/or tables related thereto, in computer readable form only, as indicated in the Supplemental Box Relating to Sequence Listing (see Section 802 of the Administrative Instructions).

4. This report contains indications relating to the following items:

- ☒ Box No. I Basis of the opinion
- ☐ Box No. II Priority
- ☐ Box No. III Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
- ☐ Box No. IV Lack of unity of invention
- ☒ Box No. V Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
- ☐ Box No. VI Certain documents cited
- ☒ Box No. VII Certain defects in the international application
- ☒ Box No. VIII Certain observations on the international application

Date of submission of the demand 15.02.2005	Date of completion of this report 28.06.2005
Name and mailing address of the International preliminary examining authority:  European Patent Office - P.B. 5818 Patentlaan 2 NL-2280 HV Rijswijk - Pays Bas Tel. +31 70 340 - 2040 Tx: 31 651 epo nl Fax: +31 70 340 - 3016	Authorized Officer Holtorf, S Telephone No. +31 70 340-2627 

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**Box No. I Basis of the report**

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1. With regard to the **language**, this report is based on the international application in the language in which it was filed, unless otherwise indicated under this item.
- ☐ This report is based on translations from the original language into the following language , which is the language of a translation furnished for the purposes of:
- ☐ international search (under Rules 12.3 and 23.1(b))
  - ☐ publication of the international application (under Rule 12.4)
  - ☐ international preliminary examination (under Rules 55.2 and/or 55.3)
2. With regard to the **elements\*** of the international application, this report is based on *(replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report):*

**Description, Pages**

1-30 as originally filed

**Claims, Numbers**

1-17 as originally filed

- ☐ a sequence listing and/or any related table(s) - see Supplemental Box Relating to Sequence Listing
3. ☐ The amendments have resulted in the cancellation of:
- ☐ the description, pages
  - ☐ the claims, Nos.
  - ☐ the drawings, sheets/figs
  - ☐ the sequence listing (*specify*):
  - ☐ any table(s) related to sequence listing (*specify*):
4. ☐ This report has been established as if (some of) the amendments annexed to this report and listed below had not been made, since they have been considered to go beyond the disclosure as filed, as indicated in the Supplemental Box (Rule 70.2(c)).
- ☐ the description, pages
  - ☐ the claims, Nos.
  - ☐ the drawings, sheets/figs
  - ☐ the sequence listing (*specify*):
  - ☐ any table(s) related to sequence listing (*specify*):

\* If item 4 applies, some or all of these sheets may be marked "superseded."

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**Box No. V Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement**

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**1. Statement**

Novelty (N)	Yes: Claims	1-17
	No: Claims	
Inventive step (IS)	Yes: Claims	
	No: Claims	1-17
Industrial applicability (IA)	Yes: Claims	1-17
	No: Claims	

**2. Citations and explanations (Rule 70.7):**

**see separate sheet**

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**Box No. VII Certain defects in the international application**

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The following defects in the form or contents of the international application have been noted:

**see separate sheet**

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**Box No. VIII Certain observations on the international application**

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The following observations on the clarity of the claims, description, and drawings or on the question whether the claims are fully supported by the description, are made:

**see separate sheet**

**Re Item V.**

**Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement.**

The following documents are referred to in this communication:

- D1 : WO 99/52345 A (PLANT BIOSCIENCE LTD) 21 October 1999 (1999-10-21)
- D2 : BRANCA FERDINANDO ET AL: "Survey of aliphatic glucosinolates in Sicilian wild and cultivated Brassicaceae" April 2002 (2002-04), PHYTOCHEMISTRY (OXFORD), VOL. 59, NR. 7, PAGE(S) 717-724 , XP002289628 ISSN: 0031-9422
- D3: WO03/004619 (Univ. California) 16 January 2003 (2003-01-16)

**Novelty (Art. 33 (2) PCT)**

Claims 1-17 do meet the requirements of Article 33 (2) PCT in that the matter for which protection is sought is new. The subject matter of independent claim 1 deals with the provision of a method for the generation of commercial Brassica oleracea plants with elevated levels of the anticarcinogenic glucosinolates Glucoiberin (3-methylsulphinylpropyl glucosinolate (3MSPG)) and/or Glucoraphanin (4-methylsulphinylbutyl glucosinolate (4MSBG)) by utilizing previously identified and selected commercial Brassica oleracea plants with an elevated level in said two glucosinolates as a starting material for breeding Brassica varieties. Such method has not been disclosed in the prior art.

**Inventive Step (Art. 33 (3) PCT)**

Document **D1 (WO9952345)**, which is considered to represent the most relevant state of the art, discloses a method for the selective increase of both anticarcinogenic glucosinolates 3MSPG and 4MSBG in edible Brassica vegetables (see page 1) by

crossing selected high-glucosinolate wild Brassica oleracea species with commercial Brassica oleracea breeding lines thereby forming hybrids between commercial and wild Brassica species.

The difference between **D1** and the subject-matter of claim 1 is the use of commercial Brassica oleracea species selected for their high 3MSPG and/or 4MSBG-content instead of wild Brassica oleracea species as the starting material for breeding of edible Brassica varieties.

The problem to be solved by the present invention may therefore be regarded as the provision of alternative Brassica oleracea species exhibiting high 3MSPG and/or 4MSBG-content as starting material for breeding.

The solution is the provision of the selected Brassica oleracea varieties *Bordeaux*, *Wirosa*, *Belstar* and *Coronado*.

The solution proposed in claims 1 of the present application can not be considered as involving an inventive step (Article 33(3) PCT) for the following reasons.

Document **D1** is already disclosing a method for the selective increase of both anticarcinogenic glucosinolates 3MSPG and/or 4MSBG in edible Brassica vegetables by crossing selected high-glucosinolate wild Brassica oleracea species with commercial Brassica oleracea breeding lines thereby forming hybrids between commercial and wild Brassica species, followed by the analysis of the F1-hybrids and the selection of those hybrids with elevated levels in said 3MSPG and 4MSBG. Faced with the identified problem of providing alternative Brassica oleracea species as starting material for breeding, the person skilled in the art would have come across the publication of B. Ferdinando, et al. , Phytochemistry (**D2**), that discloses apart from wild Brassica species, also commercial Brassica oleracea species that exhibit elevated levels in either 3MSPG or 4MSBG (see Fig. 3 and page 722, left column, third paragraph, see lines No. 33 and 27) and can thus be used as alternative starting material for the same purpose. Further analysis of the levels of 3MSPG and 4MSBG in such commercial Brassica species can be done by techniques known in the art and also used in the current application, i.e. HPLC, which is disclosed in document **D3** (see Example 1 , page 34 and Table 6).

The use of such commercial Brassica oleracea plants is straightforward because they are

even more closely related genetically with the crossing partners than the wild Brassica oleracea species, making it even easier to develop commercial Brassica oleracea varieties by plant breeding.

The claimed method consists entirely of natural phenomena such as crossing, inter-breeding or selectively breeding by bringing together those plants having certain preferred characteristics/traits. Such processes are used by the ordinary breeder and the essence of said processes is non-technical.

Therefore the features disclosed in **D1** and **D2** would be combined by the skilled person, without exercise of any inventive skills in order to solve the problem posed. The proposed solution in independent claim 1 thus cannot be considered inventive (Article 33(3) PCT). Dependent claims 2-17 do not contain any features which, in combination with the features of any claim to which they refer, meet the requirements of the PCT in respect of inventive step (Article 33(2) and (3) PCT).

**Re Item VII.**

**Certain defects in the International Application.**

**Sufficiency of Disclosure and Support (Art. 5 + 6 PCT)**

The description does not provide sufficient technical evidence to enable a skilled person to carry out the invention with respect to the subject-matter of claim 1 when interpreted in its broadest sense, i.e. extending to a method where both 3MSPG and 4MSBG are simultaneously elevated in Brassica oleracea.

The specification of the current application discloses the identification of commercial Brassica oleracea species with elevated levels in only one of the two glucosinolates 3MSPG and 4MSBG, respectively. Furthermore, there is no experimental proof provided

that such a plant can be used as a starting material for breeding commercial Brassica varieties that would exhibit elevated levels in both 3MSPG and 4MSBG. No such plant is presented.

Thus, while the breeding of Brassica varieties with only one trait (either elevated 3MSPG- or 4MSBG-content) may be easily achieved, it is considered as an undue burden in the absence of any guidance on the basis of the description or the common general knowledge to find out whether the introduction of two different specific traits (3MSPG and 4MSBG) into one Brassica variety through ordinary breeding is possible.

Consequently, the application does not fulfil the requirement of Article 5 PCT.

For the same reason, the subject-matter of claim 1 and the claims dependent thereon or referring thereto also lacks support.

#### **Essentially biological processes and plant varieties (Rule 67.1 (ii) PCT)**

The subject matter of claims 1-13 and 14-16, as long as it is supported by the description, is related to subject matter which falls under Rule 67.1 (ii) PCT.

#### **Re Item VIII**

#### **Certain observations on the International Application**

##### **Clarity (Art. 6 PCT)**

The subject matter of the independent claim 1 is not clearly defined and is open to any interpretation, extending from Brassica oleracea starting material with elevated levels in either one of 3MSPG or 4MSBG to Brassica oleracea starting material with elevated levels in both glucosinolates 3MSPG and 4MSBG or even to starting material with elevated levels

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in any other known "glucosinolate".

Furthermore, as currently drafted, the subject matter of claim 1 also comprises bred Brassica varieties with either elevated levels in only one prominent glucosinolate (3MSPG or 4MSBG) or in both (3MSPG and 4MSBG) glucosinolates at the same time. Moreover, it is not essentially clear whether the indicated concentration values of the two glucosinolates 3MSPG and 4MSBG refer to concentrations of said compounds in the edible parts of the starting material (plant as characterized under a)) or to the concentrations of said compounds in the edible parts of the Brassica varieties (see b)) which are to be generated through breeding.